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Microbiota analysis to reveal temperature abuse of fresh pork

Tasja Buschhardt[a*], Martin Iain Bahl[a], Tina Beck Hansen[a], Asker Daniel Brejnrod[b], Waleed Abu Al-Soud[b], Martin Steen Mortensen[b], Søren Johannes Sørensen[b], and Søren Aabo[a]

[a]Technical University of Denmark – National Food Institute, Kemitorvet Building 204, DK2800 Kgs. Lyngby, Denmark
[b]University of Copenhagen – Department of Biology, Section of Microbiology, Universitetsparken 15, DK-2100 København Ø

❖Introduction

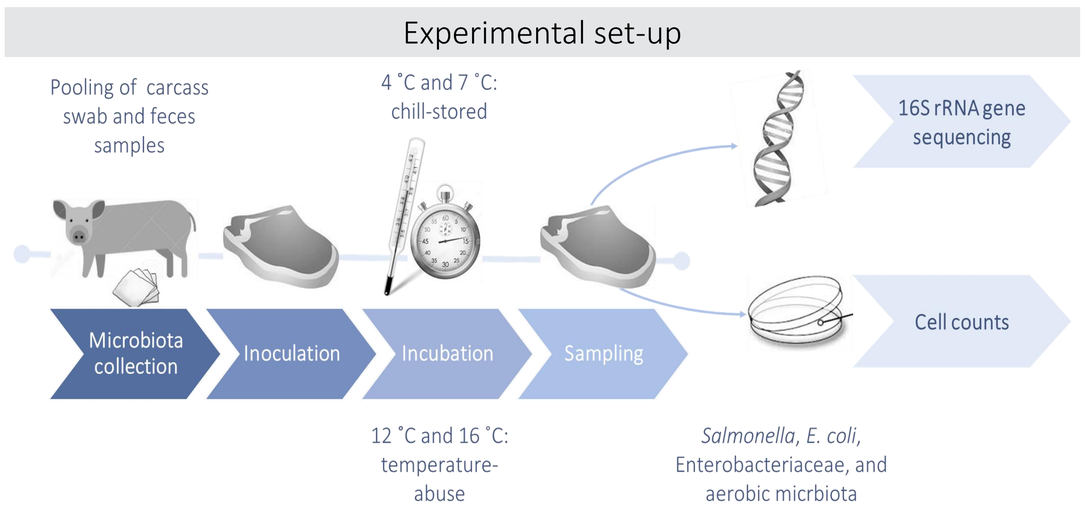
Temperature control is important to limit microbial growth along the fresh meat chain. Violations of temperature regulations may negatively affect meat safety and quality. No methods are available to estimate whether meat has been subjected to temperature abuse. Specific temperature exposures may, however, lead to systematic changes in the diverse bacterial communities associated with the meat due to different *in situ* growth characteristics. We investigated whether temperature-driven patterns in the community composition on fresh meat surfaces reflect the temperature-history (combination of time and temperature).

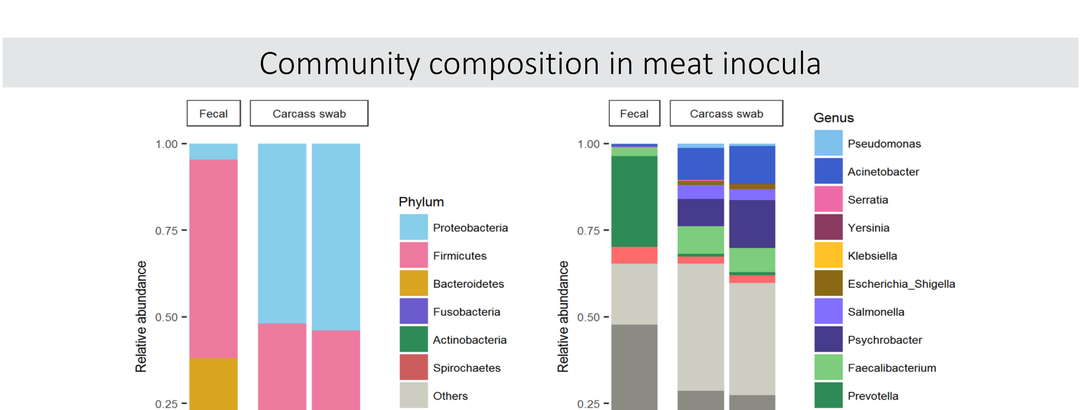
❖Conclusions & Perspectives

- PCoA of 16S rRNA gene sequencing data as a conceptual " temperature index" to indicate temperature-history (combination of time and temperature) of meat
  - Sequencing as a promising tool to study population dynamics in meat
  - Trends in quantitative data were reflected in sequencing data
  - Sequencing as a promising tool to screen for and identify growth/safety indicators
- Relative abundance data from sequencing gives valuable information but does not suffice Quantitative Microbial Risk assessment and/ or traditional quantitative testing for indicator organisms

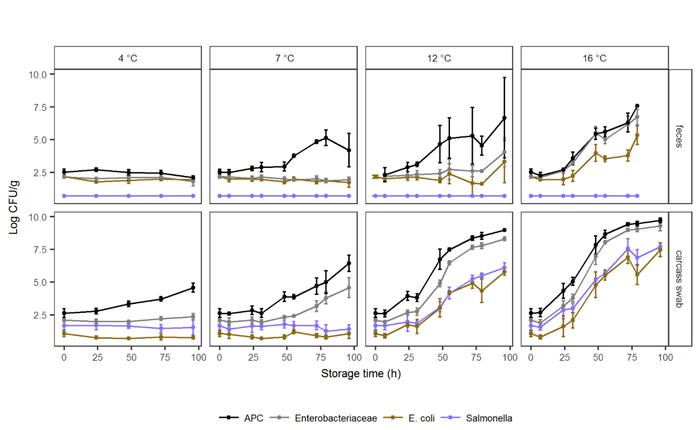
❖Methods

Sterile pieces of pork were inoculated with a carcass swab homogenate (CIM), to which *Salmonella* was added, and a fecal homogenate (FIM). Changes in the meat microbiota were monitored during aerobic chill-storage (4 °C and 7 °C) and temperature abuse (12 °C and 16 °C) for 96 hours, by culture-based methods and 16S rRNA gene sequencing.





❖Results



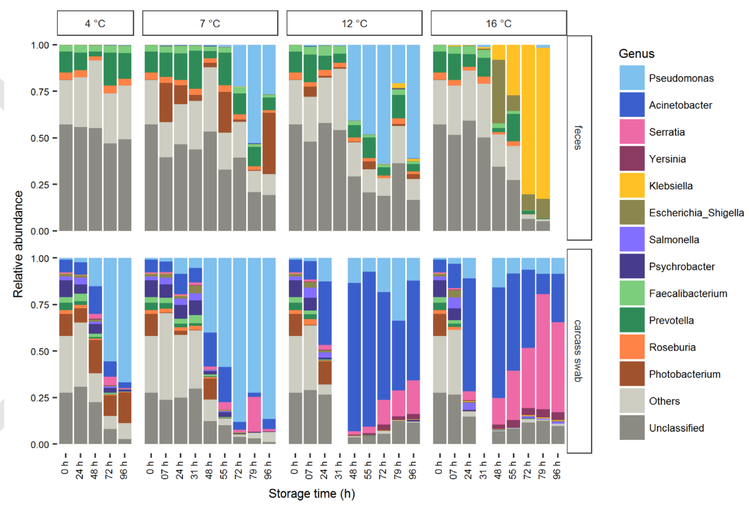
Similar growth behavior of *Salmonella* and *E. coli* on meat with carcass swab inoculum.

Observed growth of the aerobic microbiota (APC), Enterobacteriaceae, *Escherichia coli* (*E. coli*), and *Salmonella* at 4 °C, 7 °C, 12 °C, and 16 °C on FIM and CIM. Data points represent the mean log CFU/g and error bars indicate standard deviations.

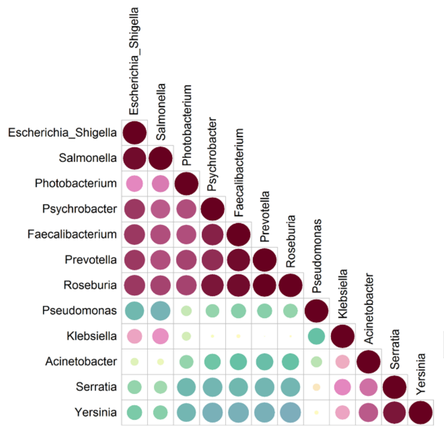
-*Acinetobacter*, *Serratia* and *Pseudomonas* dominated the meat microbiota during prolonged temperature abuse

-chill-stored meat was dominated by *Pseudomonas* only

- the composition of the initial community affects subsequent changes during storage



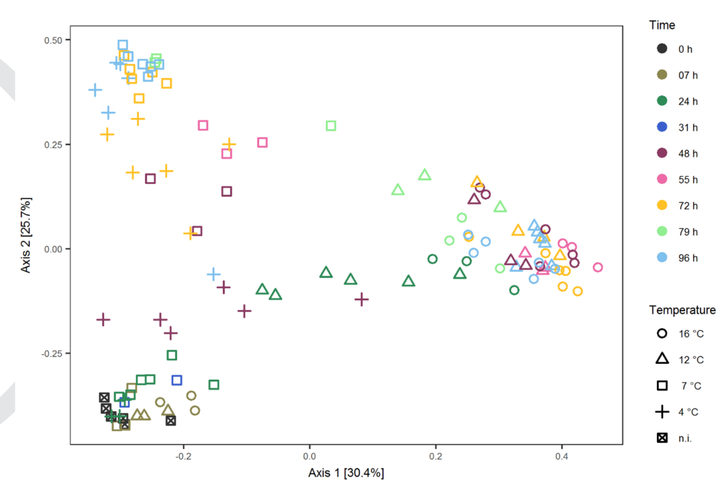
Distribution of the twelve most abundant genera during aerobic storage of FIM and CIM at 4 °C, 7 °C, 12 °C, and 16 °C. Abundances of genera with lower abundances were collectively described as *Others*.



Analysis of both sequence data and culturing revealed a strong positive correlation between *E. coli* and *Salmonella*, which suggests that *E. coli* may be used as a microbial indicator for growth of *Salmonella* on the surface of fresh pork.

Correlation matrix of the relative abundances of the twelve most abundant genera during aerobic storage of CIM at 4 °C, 7 °C, 12 °C, and 16 °C. Data from all storage temperatures and times were combined before correlations were calculated. Shading of the circles indicates the R^2-value and areas of circles show the absolute value of corresponding correlation coefficients. No circle indicates an R^2-value of zero.

The presented results suggest that principal coordinate analysis of beta diversity could be a useful tool to reveal temperature abused meat.



Changes in beta diversity during aerobic storage of and CIM at different storage temperatures. PCoA plots based on Bray-Curtis dissimilarities between storage temperature and storage times along the first two PC axes. Each point represents a single sample and is colored according to sampling times and shaped according to storage temperatures.

*busch@food.dtu.dk

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